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Post-natal exposure to corticosterone affects standard metabolic rate in the zebra finch (*Taeniopygia guttata*)

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ABSTRACT

Post-natal stress has been shown to have important short and long term effects on many adult traits in birds. During stress, metabolic alterations often result in the mobilization of energy away from energy-sensitive functions such as growth, which could have significant implications for developing animals. However, little is known about the implications of stress hormones for energy consumption in growing individuals. We experimentally increased corticosterone (CORT) levels in nestling zebra finches via oral administration, between the ages of 7 and 18 days. The standard metabolic rate (SMR) of birds was measured twice overnight when birds were between 11–13 and 55–65 days of age. Developmental CORT administration significantly elevated overnight variability in SMR (sd) in nestling birds (during the treatment period), but not at 55–65 days (5–6 weeks after the treatment period). The effect on variability was seen more prominently in birds from larger brood sizes and in females. We found no effects of our treatments on mean SMR overnight. However, brood size and sex had interactive effects, with males from larger brood sizes having higher SMR at 55–65 days of age. These results suggest that stress hormones can have significant effects on energy metabolism and possibly nocturnal arousal and sleep fragmentation. However, there were no detectable long term effects of our treatments on SMR, suggesting that these effects are only short-lived, in order to maintain homeostasis in the short term.

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1. Introduction

Stressful conditions, such as reduced food availability, promote the release of adrenal glucocorticoid hormones (Wingfield, 1994, 1998), the main such hormone in birds being corticosterone. Whilst short term increases in corticosterone are adaptive, persistent high levels resulting from continued stress can have pronounced deleterious effects (Wingfield, 1994, 1998). There is considerable evidence that elevated stress hormone levels can have significant effects on energy metabolism in adults of a wide range of animals (Astheimer et al., 1992; De Boeck et al., 2001; Laugero and Moberg, 2000a,b; Wikelski et al., 1999). During stress, changes in metabolism help support the biological defences an individual uses to maintain homeostasis (Chrousos and Gold, 1992; Wingfield et al., 1998). These metabolic alterations can result in negative energy balance via a reduction in energy-sensitive functions such as growth or body mass maintenance (Chrousos and Gold, 1992; Spencer et al., 2003; Spencer and Verhulst, 2007; Laugero and Moberg, 2000a,b). Increased stress hormone levels are also often associated with an elevation in energetically expensive behaviors

in avian species, such as foraging, a strategy thought to be beneficial in maximizing survival under stressful conditions (Astheimer et al., 1992; Breuner et al., 1998; Kitaysky et al., 2001). Whilst there is evidence that daytime metabolism may be increased, studies of nocturnal energy expenditure have produced differing results. In birds studies have suggested that experimental administration of CORT results in reduced nocturnal metabolism (Astheimer et al., 1992; Buttemer et al., 1991). This phenomenon is not manifest as a reduction in basal or minimum metabolism, but a reduction in the variation in metabolic rate experienced overnight, reducing spontaneous arousal bouts (Astheimer et al., 1992; Buttemer et al., 1991). It has been suggested that this is an adaptive strategy to reduce daily energy needs whilst under stress. In contrast, several mammalian studies have suggested that chronic stress can cause increased arousal overnight (Gronli et al., 2004; Koehl et al., 2002; Papale et al., 2005; Tiba et al., 2003).

Studies to date examining the role of stress in mediating metabolic rate have been carried out in adult animals and whilst recent evidence suggests that many species can respond to external stressors during post-natal development (Blas et al., 2005; Bowerman et al., 2002; Kitaysky et al., 1999, 2001; Love et al., 2003; Pravosudov and Kitaysky, 2006; Saino et al., 2003; Sims and Holberton, 2000; Sockman and Schwabl, 2001) there is a large gap in

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our knowledge of the energetic costs of such responses in growing individuals. In this study we aim to test the hypothesis that the metabolic response of growing individuals to elevated stress hormone levels follows the same patterns as those seen in adult birds. Kitasky et al. (Kitaysky et al., 2001) have shown that experimentally increasing corticosterone by a factor of 1.8 above basal levels in juvenile black-headed gulls resulted in significant increases in begging rate of between 60% and 180%. This was interpreted as an adaptive response to food stress by the chicks to gain access to more resources from the provisioning parents, and indeed chicks with experimentally amplified begging rates received food at higher rates, a relationship found in many species (Iacovides and Evans, 1998; Kilner, 1995; Royle et al., 2002; Saino et al., 2000). Thus growing birds exposed to corticosterone exhibit an elevated diurnal metabolism as seen in adult counterparts. Therefore if young birds follow the same energetic patterns over the whole day as adults and a dampened overnight metabolism compensates for increased diurnal activity we would predict a reduced metabolic rate overnight to compensate for this increased expenditure in animals that experience elevated levels of stress hormones.

However, all individuals may not respond in the same way to elevated stress hormones. Several studies have suggested significant sex differences in responses to developmental conditions (Arnold et al., 2007; de Kogel, 1997; Kudielka and Kirschbaum, 2005; Liu et al., 2006; Martins, 2004; Mueller and Bale, 2007; Raberg et al., 2005; Verhulst et al., 2006). Females are more susceptible to the negative effects of developmental stress in several avian species, including the zebra finch, the species used in this study (de Kogel, 1997; Martins, 2004; Verhulst et al., 2006), and there is some evidence for increased metabolic rates following stressful rearing environments (Verhulst et al., 2006). However, the mechanism underlying these sex-specific effects are still largely unknown. In addition the amount of sibling competition within a nest may restrict resources and have an additive effect under more stressful conditions. Brood size enlargements in avian species have been shown to affect growth rates, immunocompetence, body condition and long term survival (de Kogel, 1997; Deerenberg et al., 1996; Eraud et al., 2008; Naguib et al., 2004; Soma et al., 2006). Since both sex and brood size have both been shown to be important in shaping individual responses to developmental stress we would hypothesise that females and birds from large broods would be more likely to exhibit significant elevations in metabolic rate under stressful conditions.

Recently the role of early life conditions in shaping adult success and fitness has been highlighted across a wide range of species, including humans (Bateson et al., 2004; Buchanan et al., 2003; Gil, 2003; Ketterson et al., 1996; Metcalfe and Monaghan, 2001; Moussaieu and Fox, 1998; Nowicki et al., 1998; Spencer et al., 2003, 2004). In particular the experimental elevation of corticosterone during early post-natal development has been shown to influence several phenotypic traits later in life, such as sexual signals, competitive ability, cognition and fear-related behaviors (de Kloet et al., 2002; Kitaysky et al., 2003; Spencer et al., 2003; Spencer and Verhulst, 2007). One recent study has also linked early conditions, in the form of sibling competition and brood size, to metabolic rate in adulthood (Verhulst et al., 2006), however, little is known about the short term effects of sibling competition on metabolism. Thus it is possible that post-natal exposure to corticosterone could have long term effects on metabolic rate.

In this study we investigated the immediate and long term effects of experimental exposure to the stress hormone corticosterone (CORT) during the latter half of the nestling period on standard metabolic rate (SMR) and the variation in SMR in the altricial bird species the zebra finch (*Taeniopygia guttata*). The study utilized genetically related sibling pairs of birds, both male and female, from two manipulated brood sizes (three or five chicks),

where one received oral administration of small amounts of CORT between the ages of 7 and 18 days post-hatching and the other received only the carrier solution (peanut oil) during this period. This allowed us to determine the effects of sex, brood size and CORT levels on metabolism and determine any interactive effects.

2. Methods

2.1. Corticosterone manipulation

Birds in this study were also used in a previous study to determine the long term behavioral effects of stress hormones (for full details see Spencer and Verhulst, 2007). In brief, adult zebra finches were randomly paired ($n=23$ pairs) and housed at the Biological Centre at the University of Groningen in $80 \times 40 \times 40$ cm cages equipped with nestboxes and nesting material (hay). The birds were maintained at a temperature of 20–24°C and a photoperiod of 14L:10D. Breeding pairs were provided with a commercial tropical seed mixture (Teurlings, Dordrecht, The Netherlands), water, shell grit, and cuttlefish bone *ad libitum*. Pairs were also given egg food three times per week, until their offspring hatched when they received supplementary egg food daily. Upon hatching first and second hatched nestlings in each nest were individually marked by colored nail polish to each bird's toes. Brood sizes were standardized to either three or five nestlings (population range 2–6 nestlings) by adding or removing nestlings at 2 days of age (additional nestlings in a nest were aged within 1 day of foster siblings), ensuring that two siblings (first and second hatched) remained in each nest that were genetically related to the parents. This was done to determine any effects of brood size on SMR parameters. When nestlings reached 7 days of age experimental treatments began. First and second hatched chicks from each breeding pair were assigned to one of two treatments: corticosterone administration or control, counterbalancing across broods for chick hatch order to control for any potential differences in basal corticosterone levels (Schwabl, 1999). As described in Spencer and Verhulst (2007) birds in the corticosterone group were dosed with 0.0125 mg of corticosterone daily, dissolved in peanut oil (concentration 0.25 mg/ml) via two 25 μ l doses (at least 6 hours apart) from the age of 7–18 days of age. This dosing schedule results in a short term increase in plasma corticosterone levels (Spencer and Verhulst, 2007). To determine this single blood samples were taken a sub-sample of birds ($n=32$) at different times following dosing on day 12 post-hatching (details in Spencer and Verhulst, 2007). Following dosing CORT levels peaked after 10 min and the levels of CORT experienced by our nestlings fell within the natural range of nestling zebra finch responses to a standardized capture handling restraint protocol at 16 days post-hatching (Wada et al., 2008) and we have recently collected data to suggest that the peak response to a standardized stressor does not differ between nestlings of 12 and 16 days of age (Spencer et al., unpublished data). The sampling regime used to determine the effects of our treatment meant that a sub-sample of birds ($n=16$) had been bled the day before our metabolic measurements, however, all siblings were measured on the same day and these birds are spread equally across sex and brood size. Control birds were dosed with the same amount of the peanut oil carrier solution at the same time as their sibling. All manipulations ceased at the age of 18 days. All birds had their mass recorded at regular intervals, particularly during each metabolic rate measurement at day 12 and 55–65 days. The sex of each bird was determined via the presence of sexually dimorphic adult plumage, which begins to appear after 35 days of age. Due to mortality two birds were removed from the analysis, leaving us with a final sample size of 44 birds (from 23 sibling pairs); a further breakdown of sample sizes can be seen on the relevant figures. All procedures were carried out following peer review and approval from the ethical committee for animal experiments of the University of Groningen (under license D4299).

2.2. Metabolic rate measurements

Metabolic measurements were carried out as described in Verhulst et al. (2005). Briefly, each bird was removed from its nest between 18.30 and 19.30h, 1.5–2 h prior to the end of the light period and placed inside a dark 2-l Plexiglas box equipped with a perch at a temperature representative of the prevailing ambient temperature. At 11–13 days of age this meant maintaining a temperature of 30 °C, the mean temperature within the nest cup. The second trial was conducted when birds were between 55 and 65 days of age, hence the temperature was set at 22 °C to mimic their holding cage conditions. Although the age of birds varied in the metabolic trials across sibling pairs, each pair of birds was measured on the same night to control for potential age effects. The air flow rate was controlled by mass-flow controllers (5850S, Brooks, Rijswijk, The Netherlands), that were calibrated with a bubble flow meter, set to deliver 16 l·h⁻¹. In- and out-flowing air was dried by passing through a molecular sieve (3 Å, Merck, Darmstadt, Germany). Gas analysis was done using a paramagnetic O₂-analyser (Servomex Xentra 4100, Crowborough, UK) and CO₂-analyser (Servomex 1440). The system was calibrated before each measurement session using two three-digit precision gas mixtures of 20.0% O₂/0% CO₂ and 21.0% O₂/1.0% CO₂ in N₂. CO₂ and O₂ measurements were recorded at 7 min intervals (between 81 and 87 recordings were taken over the measurement period) for approximately 10 h, which is the normal length of the dark period experienced by these birds in this experiment. The rate of oxygen consumption was calculated from these measurements and converted to the energy equivalent, while correcting for the respiratory quotient, according to Brody (Brody, 1945). SMR was taken to be the minimum value of a 30 min running mean for each individual. In addition the standard deviation of the minimum 30 min running mean was also calculated to determine variability in nocturnal energy expenditure (here called standard deviation of SMR), in line with Buttemer et al. (1991). Body mass was measured before and after respirometry.

2.3. Statistical analysis

All data were analyzed using Linear Mixed Models (SPSS, version 13.0), with cage of birth entered as random effect. SMR and the standard deviation of SMR were used as dependent variables in each analysis. For all analyses data from the two trials were combined to perform repeated-measures models with individual nested within cage of birth entered as an additional random factor. Fixed variables in each model were: brood size (three or five), sex, treatment (control or CORT), age (11–13 or 55–65 days) and hatching order (first or second). In addition mass (mean of values taken before and after each respirometry measurement) was entered as a covariate. Stepwise deletion of non-significant non-hierarchical terms was employed. All residuals were checked for normality.

3. Results

3.1. Nocturnal metabolism

The analysis of SMR showed a significant interaction between age, sex and brood size ($F_{1,55.3}=6.20$, $p=0.015$), where females from both brood sizes and males from small brood sizes (containing three chicks) showed similar small decreases in SMR with age, but in males from large brood sizes (containing five chicks) SMR was maintained over time (Fig. 1). There was also a significant main effect of age ($F_{1,34.1}=4.62$, $p=0.034$), indicating in this case that the intercepts of the two metabolic trials differ significantly. Birds from second hatched eggs also exhibited significantly elevated metabolic rates, independent of mass as the model controlled for this, showing an increase of 6% over birds from first hatched eggs

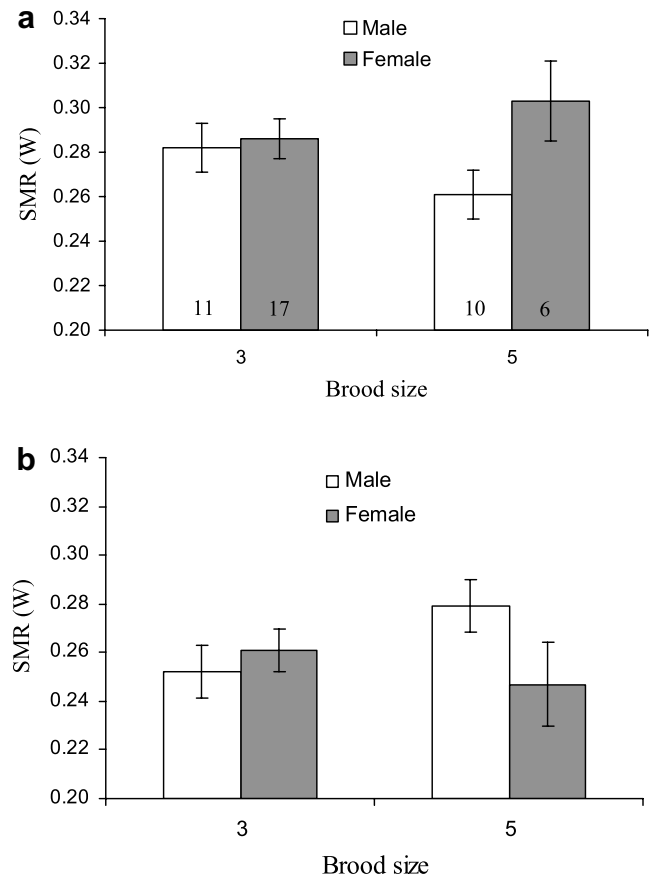


Fig. 1. The effect of manipulated brood size on the standard metabolic rates (W) of male and female zebra finches measured at (a) 11–13 and (b) 55–65 days post-hatching: age \times brood size \times sex interaction: $F_{1,55.3}=6.20$, $p=0.015$. Sample sizes are shown within each bar.

($F_{1,36.9}=4.62$, $p=0.036$). Mean mass was significantly positively related to SMR ($F_{1,33.3}=31.7$, $p<0.0001$). However, there were no effects of treatment on SMR and no other significant interactions ($p>0.4$). There were no differences in the masses of siblings at either 11–13 or 55–65 days of age ($F_{1,40}=0.34$, $p=0.56$, $F_{1,40}=0.03$, $p=0.81$, respectively), however, birds fed with CORT in this population did show reduced mass compared to controls at the end of the treatment period (18 days of age, $F_{1,18.5}=31.91$, $p<0.001$, statistics from Spencer and Verhulst, 2007).

3.2. Variation in SMR

There was a significant interaction between age and treatment ($F_{1,36.9}=5.27$, $p=0.03$; Fig. 2), as CORT administration had immediate effects on the variability of nocturnal metabolism but no long term effects, with CORT-fed birds showing a trend towards elevated standard deviations of SMR during the nestling period ($t_{(21)}=1.4$, $P=0.08$). Brood size also interacted with treatment and age ($F_{1,37.9}=4.82$, $p=0.03$, Fig. 3), suggesting that the interaction between age and treatment was driven by the effects seen in larger brood sizes, whilst those birds from small brood sizes show similar responses to the treatment. Again there was a significant main effect of age ($F_{1,36.9}=20.7$, $p<0.0001$), and a non-significant trend towards an effect of brood size ($F_{1,22.9}=3.44$, $p=0.07$). There was also a significant interaction between sex and treatment ($F_{1,38.7}=7.15$, $p=0.011$; Fig. 4), with CORT-fed females showing significantly more variability in SMR ($t_{39}=2.0$, $p=0.05$), whilst males showed no metabolic responses to CORT administration ($t_{39}=1.1$, $p=0.27$). There was also a non-significant trend towards a three-

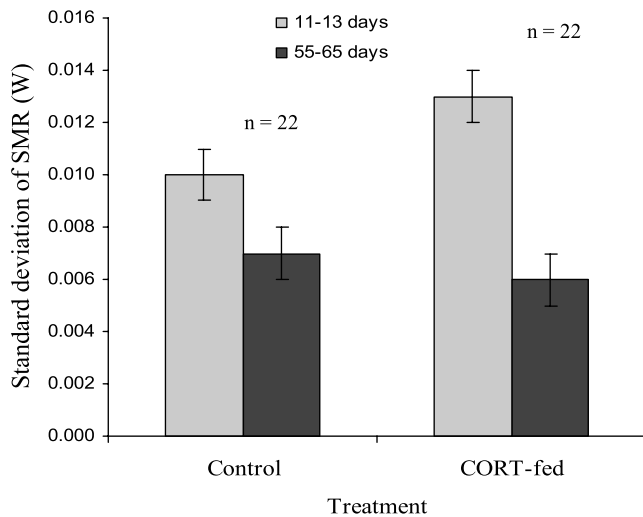


Fig. 2. The effects of age and CORT treatment on the standard deviation in SMR (W), suggesting that stress hormones administered during post-natal development have only short term effects on metabolism. Treatment \times age interaction: $F_{1,36.9}=5.27$, $p=0.027$. Sample sizes are shown above treatment.

term interaction (age \times treatment \times sex interaction: $F_{1,38.1}=3.86$, $p=0.056$), confirming that these treatment effects are mainly due to changes in female metabolism during the trial at 12 days of age. There were no effects of hatching order on SMR variability ($F_{1,37.4}=0.26$, $p=0.63$).

4. Discussion

4.1. Short term effects on metabolism

Our results demonstrate that developmental exposure of avian nestlings to glucocorticoid hormones can have significant short term effects on standard metabolism, resulting in increased variability overnight. Whilst previous work has provided evidence for metabolic changes in response to stress in adults, to our knowledge this is the first study to examine this directly in growing individuals. This increase in metabolic variability is contrary to our hypothesis that we would find the same relationship found in adult birds, i.e. that elevated CORT levels decrease nocturnal metabolism by reducing the amount of arousal events overnight (Astheimer et al., 1992; Buttemer et al., 1991). It has been suggested that this regulates daily energy budgets and re-coups reserves for the following day in order to avoid or overcome the stressor (Astheimer et al., 1992; Buttemer et al., 1991). Our data suggest that growing birds may be more sensitive to stress than adults, or may be less able to regulate their energy expenditure over a 24 h period. Our results are, however, more consistent with work on mammals, that suggests chronic stress causes increased nocturnal arousals, i.e. increased variability in metabolic rate, which have been linked to increased sleep fragmentation (Gronli et al., 2004; Koehl et al., 2002; Papale et al., 2005; Tiba et al., 2003). Such sleep disturbances are known to increase overnight energy expenditure, allostatic load and energy imbalance (Bonnet et al., 1991; McEwen, 2006). We did not measure sleep fragmentation in this study and therefore can draw no firm conclusions about the effects of our treatments on sleep parameters and further work would be required to determine this. Although birds from the CORT-fed group in our study did not show reduced masses at the time of the metabolic measurements, body mass in these birds was significantly reduced at the end of the treatment period, at 18 days of age (Spencer and

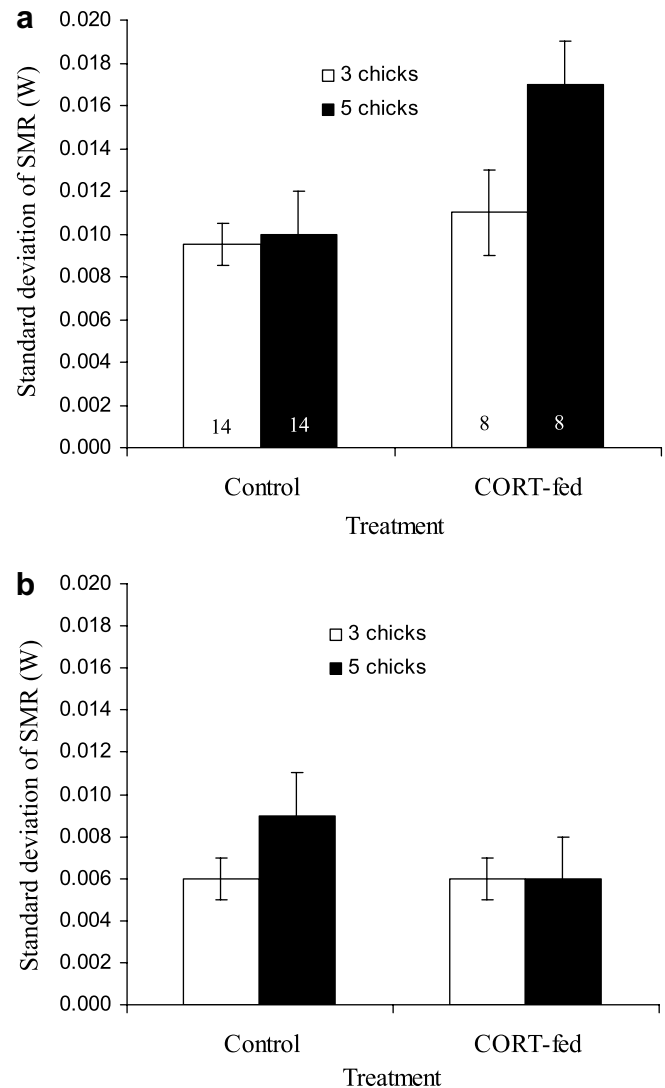


Fig. 3. The effect of CORT treatment and brood size ($n=3$ or 5 nestlings) on the variation in SMR (W) during two trials at age (a) 11–13 and (b) 55–65 days: treatment \times age \times brood size interaction: $F_{1,37.9}=4.82$, $p=0.03$. Sample sizes are shown within each bar.

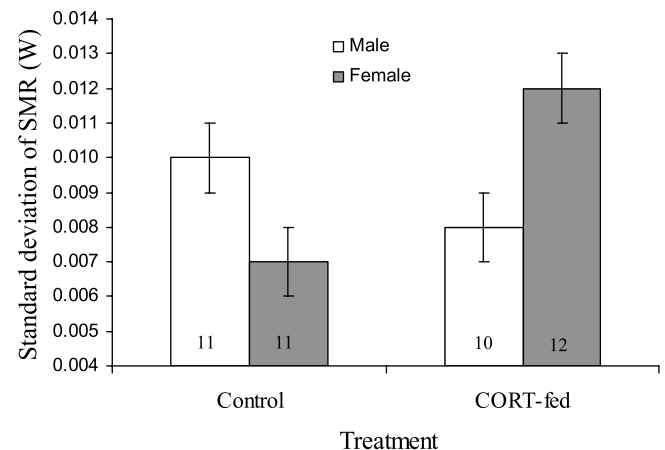


Fig. 4. The effect of CORT administration on the variability of SMR in male and female zebra finches. Treatment \times sex interaction: $F_{1,38.7}=7.15$, $p=0.011$. Females showed a significant increase in variability following CORT treatment ($t_{39}=2.0$, $p=0.05$), whereas males showed no response ($t_{39}=1.1$, $p=0.27$). Sample sizes are shown within each bar.

Verhulst, 2007). This suggests that repeated stress for a prolonged period may have an incremental effect on growth.

Our experimental manipulation mimics a natural increase in glucocorticoids following a stressor. This repeated increase in stress hormones would have the increased allostatic load of the individual, i.e. the cumulative costs of maintaining the internal stability of the animal (Korte et al., 2005; McEwen, 2006; Wingfield, 2005). The observed responses to the elevation in stress hormones could then be part of an allostatic response to stressful conditions to reduce overall allostatic load. However, it is difficult to see how an increase in metabolic variability and potentially overnight arousal would confer any metabolic advantage on the animal and restore the animal to its normal state. It is more likely that the stressor imposed by our manipulations resulted in allostatic overload (type 2), which did not trigger transfer to an emergency life history stage, but instead conferred significant constraints on the individual, depressing growth rates (Korte et al., 2005; Wingfield, 2005). However, further work is required to identify the possible adaptive significance of the metabolic changes seen in this study.

Exposure to CORT did not affect all individuals in this study. We found that CORT-fed individuals from experimentally enlarged brood sizes had significantly more variability in SMR. Brood size enlargements in this species have been shown to affect growth rates (de Kogel, 1997; Deerenberg et al., 1996), and produce long term effects on survival post-independence (de Kogel, 1997; Deerenberg et al., 1996). Sibling rivalry would have been more intense in these broods, along with a possible reduction in food intake. Thus the results suggest that there are additive effects of 'stress' at work here, and are in line with our prediction that brood size would show an interaction with elevated stress hormones. It is possible that birds in larger broods had elevated CORT levels, which might have augmented our experimental procedures, but we have no data to evaluate this hypothesis. Our results suggest that birds in large broods would suffer larger metabolic changes during periods of further food shortages or another external stressor than birds in small broods.

We also detected sex differences in the metabolic response to glucocorticoid exposure, as predicted. Although there is no sexual size dimorphism in zebra finches, previous studies have suggested that females may be more susceptible to nutritional stress through either food restriction or brood size manipulation during development, resulting in decreased survival or altered metabolism (de Kogel, 1997; Martins, 2004; Verhulst et al., 2006). Interestingly Martins (2004) showed that females grew more slowly than males when under identical experimental food restriction in the absence of sibling competition. Since our results suggest that females do not have a damped nocturnal energy strategy to compensate for any increases in daytime activity, such as begging for food, this may provide a mechanism by which growing females under chronic stress show reduced growth rates. The fact that males in this study did not seem to respond to the experimental treatment, but were more sensitive to the amount of sibling competition during the nestling stage (brood size), may suggest sex-specific mechanisms for coping with stressful environments.

Previous studies in birds that have utilized experimental manipulations of glucocorticoids have mainly used methodologies that cause elevations in baseline levels of CORT, such as silastic implants (Astheimer et al., 1992; Buttemer et al., 1991). These techniques would mean that baseline levels were probably elevated over the whole 24 h period, including the nocturnal period when metabolic measurements were taken. In contrast, our feeding regime only caused short term increases in CORT for less than 2 h after dosing (Spencer and Verhulst, 2007). This difference in methodologies may explain the differences seen in overnight metabolism. However, we assert our experimental procedure is biologically more relevant when aiming to mimic repeated acute stressors, such as

predator attacks or disturbance, in particular because strong fluctuations in plasma levels are inherent to glucocorticoid hormones. However, we still know very little about nestling responses to such natural stressors and more work is required in this area. One study in a laboratory population of rats has suggested that acute stressors applied at either the onset or conclusion of the light period always show their effects on sleep fragmentation within the dark cycle (Koehl et al., 2002). This confirms that CORT can have delayed effects on metabolism and sleep and supports our interpretation of the current results.

4.2. Long term effects on metabolism

We found no long term effects of experimental CORT administration on the variability of SMR. However, SMR tended to decrease over the two respirometry trials in all birds from small brood sizes and in females from larger broods, despite a decrease in ambient temperature, which would normally be associated with an increase in metabolic rate (Gavrilov, 1996). Whilst several studies have shown that resting metabolism is highly repeatable in avian species (Bech et al., 1999; Ronning et al., 2005; Vezina and Williams, 2005), they have not yet compared metabolism during the nestling stage with later stages of development. In this study SMR comparisons between the two metabolic trials revealed non-significant repeatability in both of our metabolic parameters (SMR: $F_{43,44} = 0.84$, $p = 0.72$; sd : $F_{43,44} = 1.17$, $p = 0.30$). An age-related decline in standard or resting metabolism has, however, been well documented in mammalian species, including humans (Hughes et al., 1998; Neuhauser-Berthold et al., 2000; Speakman et al., 2003; Van Pelt et al., 1998, 2001) and is thought to relate to changes in body composition (Hughes et al., 1998). However, these studies tend to measure age-related changes during adulthood and this negative relationship is not universal. In one recent avian study has provided evidence for a positive relationship between age and resting metabolic rate (Pis and Lusnia, 2005).

In addition we found a significant effect of hatching order on SMR, regardless of age and independent of mass, with second hatched chicks exhibiting elevated metabolic rates compared to their first hatched siblings. Previous work in kestrels has suggested that hatching order can affect metabolism, with first hatched chicks showing elevated daily energy expenditures (Massemin et al., 2003) and last hatched chicks showing significantly lower resting metabolism compared to their older siblings (Massemin et al., 2002), which are at odds with our results. However, first and second hatched chicks have been shown to have differential levels of both baseline and stress-induced CORT during the nestling period, invariably with first hatched chicks showing elevated hormone levels (Schwabl, 1999; Love et al., 2003). Therefore endogenous levels of CORT may differ across hatching order in our population and play a role in mediating SMR both during the nestling period and in later life. However, it is likely that these birds differ in several other aspects and further work would be required to elucidate the mechanisms underlying the effects seen in this study.

There were also persistent effects of brood size on SMR, as males from nests with a brood size of five failed to show the age-related decline in metabolic rate seen in other birds in this and other studies (Hack, 1997; Klausen et al., 1997), resulting in elevated rates in some males at sexual maturity. A recent study using the zebra finch has also shown long term effects of brood size on standard metabolic rate (Verhulst et al., 2006); however, in contrast to our findings they reported stronger effects on metabolism in female birds. Very similar methodologies were used to quantify SMR in these two studies, but in the earlier study birds were measured when they were over one year old. However, both studies suggest that sibling competition can have long term effects on metabolism in later life.

In summary, our results show that exposure to glucocorticoid hormones can have significant effects on the variability of nocturnal metabolism, specifically in broods that had experienced heightened sibling competition. This could have important consequences for energy balance and growth over the nestling period, however, the true fitness costs of such changes to metabolic rate require further investigation. Whilst we found no long term effects of our experimental treatments, our results also suggest that increased sibling competition can elevate standard metabolism in the long term.

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